# *Contribution*[1]

*iGEM 2022, University of Sheffield*

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Making foundational advances in bringing directed evolution to everyone everywhere

As an iGEM team in the Foundational Advance track, our aim is to make a helpful contribution to future iGEM teams. With this in mind, our ultimate motivation for rEvolver was to make directed evolution a feasible means of protein optimisation for smaller labs with finite resources and time. In facilitating automated protein evolution, rEvolver allows users to devote their time to other areas of their work - pushing forward what is achievable in the time constraints of a summer project. Alongside this, we have developed tools to complement rEvolver: an affordable, easy to assemble toroidal bioreactor, and our ultimate bioinformatics toolbox with staple molecular biology tools combined in one sleek user interface. Although these tools assist in vivo directed evolution, it is important to note both the bioreactor and the toolbox are intended for all projects requiring the maintenance of continuous culture, or the use of molecular cloning tools. Hence, in the spirit of iGEM, as an interdisciplinary, global competition built upon the tenets of open science - we are excited to see all the weird and wonderful iGEM projects that can benefit from the diverse range of tools we have developed.

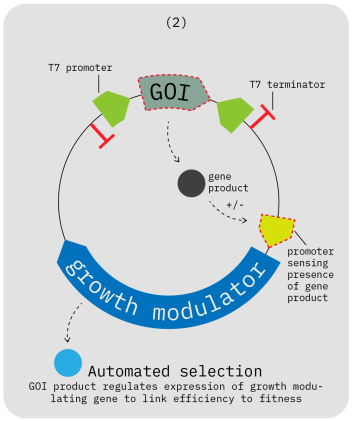
## *modular plasmid design*

rEvolver allows automated continuous in vivo directed evolution for any gene that has a complementary biosensor. The biosensor regulates the expression of a growth modulating gene to link increased efficiency to growth advantage, removing the labour intensive, repetitive steps of directed evolution. To allow the seamless integration of rEvolver into a range of projects, we have designed our plasmids to prioritise modularity. In the figure above, the red dashed lines surrounding the GOI and biosensor indicate areas that other iGEM teams could clone in their desired GOI / biosensor pair with standard molecular cloning protocols.[2] To allow further integration of rEvolver into future projects, we have made the plasmid maps readily available so that future iGEM teams can adapt our design to their heart's content. For further detail of the mechanisms of rEvolver, and how we plan the end user to implement this system, please visit our Project Description [3] and Implementation pages [4]

## *Accessible Bioreactor Design and Construction Manual*

Our toroidal turbidostat has been an overall success. It is composed entirely of affordable, widely available materials and components,

[1] from https://2022.igem.wiki/sheffield/contribution




[2]

[3] https://2022.igem.wiki/sheffield/description

[4] https://2022.igem.wiki/sheffield/implementation

has a method of construction that can be completed with your regular garage tool box and is programmed using Python — an open-source and easy-to-learn programming language. The code is thoroughly commented for ease of use and understanding, assisting those with minimal programming experience in making meaningful changes to suit their needs. It is also hugely customisable and modular. Without the OD measurement system it can still function as a regular chemostat, and with the addition of LEDs of appropriate intensity and wavelengths it can be converted into a photobioreactor.

Alongside our bioreactor, we have created an easy to follow, fully-costed construction manual[5] to assist future users in understanding how they could themselves construct one. This manual can also be used as inspiration for future teams when writing their own construction manuals for their own bioreactors that may or may not be based upon ours. Hence, in the spirit of contributing back to the iGEM community, our bioreactor could be used in a wide range of future projects of all budgets and skill levels — enabling them to run experiments that would not have been possible without a bioreactor.

[5] https://static.igem.wiki/teams/4451/wiki/bioreactor/bioreactor-construction-manual.pdf

## *A Bioinformatics Toolbox for All*

The bioinformatics toolbox has been designed to find a good balance between user friendliness and versatility. It was designed primarily to unify many of the staple tools that geneticists and synthetic biologists routinely use and streamline them into a common, navigable interface. You can find the code and links to a live demo here[6]

[6] https://gitlab.igem.org/2022/software-tools/sheffield

Currently our web-based toolbox is capable of performing the following functions upon a given DNA, RNA, or amino acid sequence:

- Determining length, GC content and sequence type of all inputs
- Converting between upper and lower case
- Reversing the sequence
- Counting element frequencies (bases or residues)
- Reverse complementing DNA and RNA
- Converting between DNA, RNA and amino acid sequences
- Finding Open Reading Frames (ORFs)

Additional tools that have been developed and incorporated in the underlying Rust library are:

- Subsequence isolation

- Calculating Hamming and Levenshtein distances

We feel that this is an impressive array of functionality for a toolbox that also features a clean, modern and intuitive UI. We know that future Sheffield iGEM iterations will definitely use it going forward, and we feel confident that our toolbox will become a favorite for many others outside of our uni!

Finally, since our toolbox is developed on GitHub in a completely open-source manner, any future users of our toolbox could contribute tools and improvements of their own. In this way, we can guarantee that our software tool will continue to grow and evolve independent of our input — a contribution that genuinely belongs to the community.